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SEXING *HYLURGOPINUS RUFIPES* (EICHHOFF) (COLEOPTERA: SCOLYTIDAE) WITH SCANNING ELECTRON MICROSCOPY

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Abstract.—Morphological structures including the size and shape of antennal clubs, the presence of ostioles on female antennal clubs and membranous prothoracic cavities on females are described. When viewed with SEM, these previously undescribed characteristics can provide positive sexual identification of *H. rufipes.*

Few scanning electron microscopy (SEM) studies have been conducted in order to describe antennal morphology of scolytid species (Payne et al., 1973; Borg and Norris, 1971) and bioacoustic mechanisms (Barr, 1969; Michael and Rudinsky, 1972; Rudinsky and Michael, 1973). Except for taxonomic illustrations by Bright (1976) there are no published reports of SEM having been utilized to study *Hylurgopinus rufipes* (Eichhoff), a major vector of Dutch elm disease in northern sections of the United States and southern Canada. To effectively diminish this beetle's role as a vector, knowledge of beetle-beetle and beetle-host relationships is essential. SEM studies can augment this knowledge.

The studies, referred to above, were performed with species for which pheromones or aggregation attractants are known to exist. However, Gardiner (1979) reported that there is no evidence for pheromone production by *H. rufipes.* As research continues on *H. rufipes* in relation to the existence of chemical cues (J. W. Peacock, personal communication), the ability to accurately sex the insect for bioassay purposes is essential. No externally identifiable sex characters have been reported for *H. rufipes* (Kaston, 1936). Our investigation described antennal morphology and a heretofore undescribed secondary sexual characteristic. Both can distinguish the sexes of *H. rufipes* when viewed with an SEM.

MATERIALS AND METHODS

Sexing involved examining the terminal abdominal segments for movement. Rapid movement of these segments indicated a male, whereas lack of movement or slow movement indicated a female. Utilizing the above behavioral trait, 50 *H. rufipes* of each sex were selected for SEM studies. Dissections have proven this method to be 90% accurate (Lanier, unpublished data).

After sexing, specimens were mounted on aluminum stubs with conductive cement and sputter-coated (Model Hummer V, Technics[1], Springfield, Va.) with

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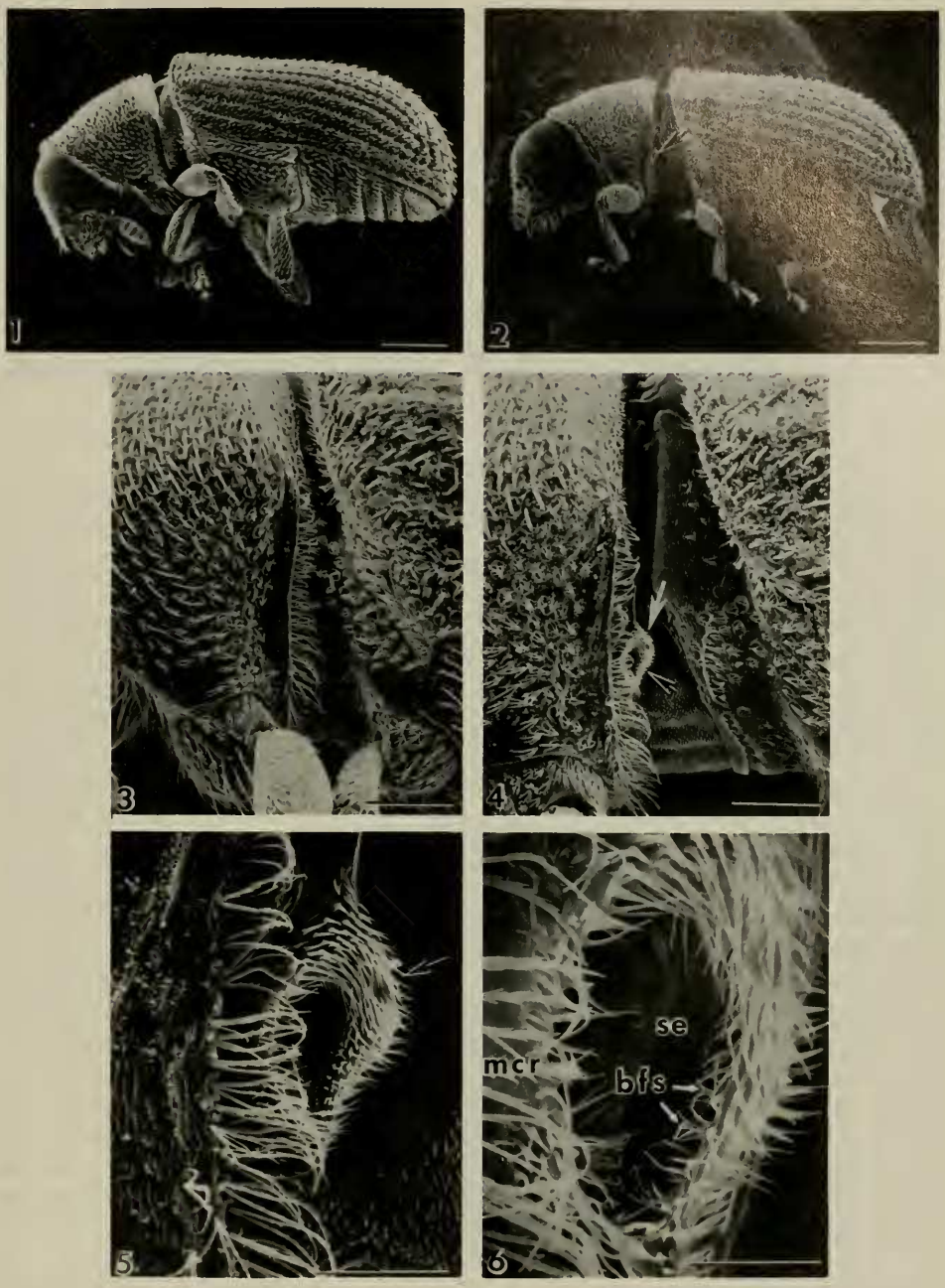


Figs. 1–6. *Hylurgopinus rufipes*. 1, Adult male (bar = 500 μm). 2, Adult female (note membraneous cavity) (bar = 500 μm). 3, Posterior margin of prothorax of male, lacking a cavity (bar = 140 μm). 4, Cavity on posterior margin of female prothorax (bar = 140 μm). 5, Globular matrix (arrow) on setal border of cavity in female (bar = 50 μm). 6, Mechanoreceptors, bifurcated setae and simple setae visible in and around cavity of female (bar = 20 μm). Abbreviations: bfs = bifurcate setae; mcr = mechanoreceptor; se = simple setae.

Table 1. *Hylurgopinus rufipes* antennal club parameters (μm) ($n = 50$).

	Length		Width	
	Mean	Range	Mean	Range
Female	250	240–265	160	155–170
Male	300	296–320	120	110–125

500 Å of Au, SEM observations were performed with a Hitachi Model S-500 (Mountainview, Calif.) at 20 kV accelerating voltage. Verifications of secondary electron images were performed with a light microscope (Zeiss IV-B, New York, N.Y.). Ten males and ten females were dissected after SEM observation to confirm sex.

Results and Discussion

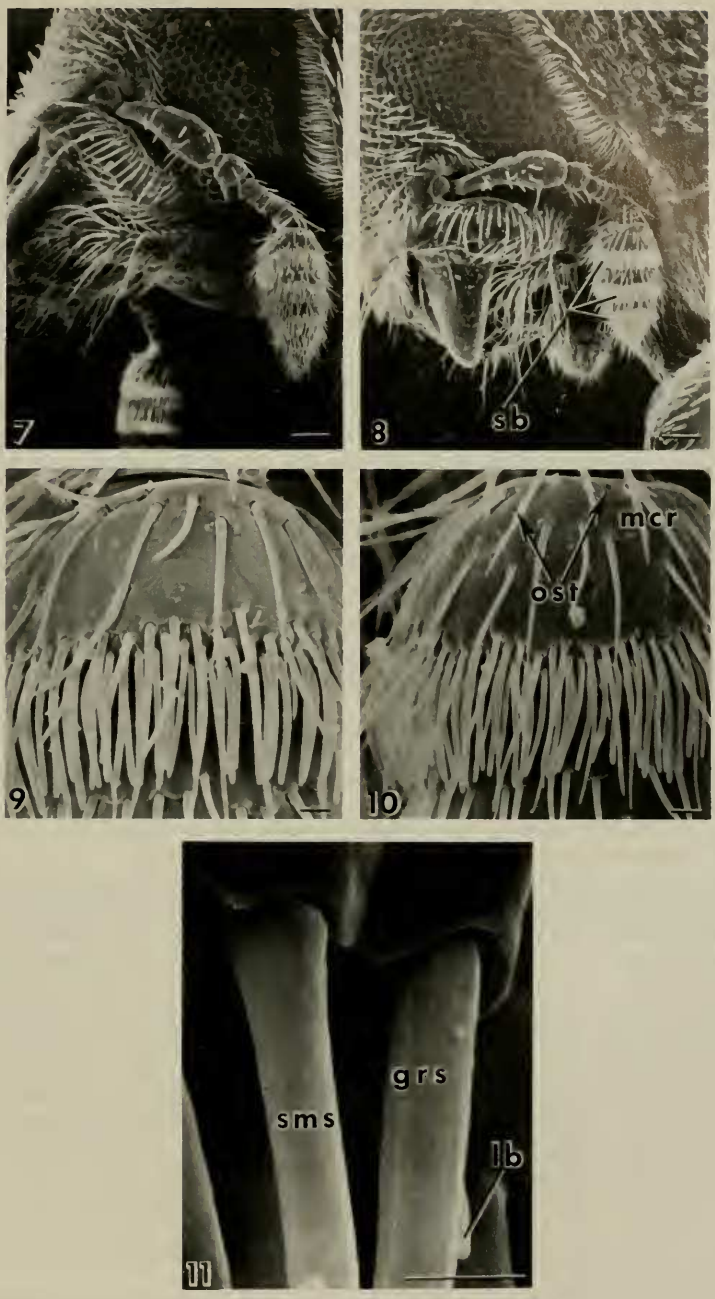
Lateral views are shown of a *H. rufipes* male (Fig. 1) and female (Fig. 2). A membranous cavity surrounded by a dense setal border can be observed on both sides of the lateral posterior region of the prothorax of females (Figs. 2, 4, 5, 6). The cavity was visible with SEM (34X) on gold coated specimens, but was not observed with a light microscope on uncoated beetles. The cavity was positioned under the posterior margin of the prothorax and could not be evaginated. The cavity was not observed on males (Figs. 1, 3). The cavity and associated setae measured ca. 100 μm × 50 μm. Setae were present on the cavity floor (Fig. 6). A globular matrix was seen within the setal border (Figs. 5, 6) on at least ten beetles.

Since serial sections were not made of the female prothoracic area associated with the cavity, we cannot definitively relate the structure to function. Further studies are needed to determine if the cavity is a simple invagination or an opening of a secretory duct. The globular matrix observed within setal borders suggests a glandular function. Faustini (1980) found similar structures (which he referred to as setiferous sex patches) on numerous beetle species. In several instances, Faustini found that setiferous sex patches were responsible for the release of pheromones.

The antenna of *H. rufipes* consists of a seven-segmented funicle and a club that varies in size and shape according to sex. The male antennal club is significantly ($P < .05$) longer and more protracted than the female antennal club (Table 1, Figs. 7, 8). Antennal clubs of both sexes have three distinct sensory bands, with the distal band being the largest. Within the sensory bands were numerous sensilla. The types and sizes were nearly identical in both sexes. A notable sex-related difference was the presence of ostioles on the proximal end of female antennal clubs (Fig. 10).

Table 2. Length of *Hylurgopinus rufipes* antennal sensilla (μm).

	Mean	Range
Smooth	14	13–17
Grooved	22	20–24



Figs. 7–11. *Hylurgopinus rufipes*. 7, Antenna of male, longer and more protracted than female antennal club (bar = 80 μm). 8, Antenna of female (note sensory bands) (bar = 80 μm). 9, Ostioles absent from proximal end of male antennal club (bar = 4 μm). 10, proximal end of female antennal club showing ostioles and mechanoreceptors (bar = 4 μm). 11, Smooth and grooved sensilla found on antennal clubs of both sexes (bar = 3 μm). Abbreviations: grs = grooved sensilla; lb = lateral branch; mcr = mechanoreceptor; ost = ostia; sb = sensory bands; sms = smooth sensilla.

Smooth and grooved sensilla were found within the sensory bands (Fig. 11). Numbers of each sensillum were not determined. Grooved sensilla were considerably longer (Table 2) and had short lateral branches present (Fig. 11). On a given sensillum the number of lateral branches were few but counts were not obtained.

Smooth sensilla were enlarged at the point of contact with their sockets (Fig. 11). These sensilla are similar to sensilla basiconica of other scolytids (Payne et al., 1973). Electrophysiological studies with sensilla basiconica on other insects have demonstrated that sensilla may be responsive to pheromones (Kinzer et al., 1969; Silverstein et al., 1968).

Mechanoreceptors were present on the funicle and none were observed on the club (Fig. 9, 10). Mechanoreceptors are often found "protecting" an underlying band of sensory sensilla, but as is the case with *Dendroctonus* spp., the smooth sensilla on *H. rufipes* lie flat and may not require protection (Payne et al., 1973).

In addition to behavioral traits used previously, we now know of several morphological structures by which *H. rufipes* can be sexed. These include the size and shape of antennal clubs, the presence of ostioles on female antennal clubs and a membranous cavity on females.

Contrary to findings of Gardiner (1979), our findings concerning the types of antennal sensilla and the presence of what may be a secretory duct indicate that some form of chemical communication system may be utilized by *H. rufipes*.

Literature Cited

Barr, B. A. 1969. Sound production in Scolytidae (Coleoptera) with emphasis on the genus *Ips*. Can. Entomol. 101: 636–672.

Borg, T. K. and D. M. Norris. 1971. Ultrastructure of sensory receptors on the antennae of Scolytus multistriatus. Z. Zellforsch. Mikrosk. Anat. 113: 13–28.

Bright, D. E. Jr. 1976. The insects and arachnids of Canada, Part 2. The bark beetles of Canada and Alaska. Coleoptera: Scolytidae. Can. Dep. Agric. Publ. 1576, 241 pp.

Faustini, D. L. 1980. Characterization of an aggregation pheromone and its site of production in *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae) with comparative notes on analogous structures in other Coleoptera families. Ph.D. Dissertation, The Ohio State University, Columbus, Ohio.

Gardiner, L. M. 1979. Attraction of *Hylurgopinus rufipes* to cacodylic acid-treated trees. Bull. Entomol. Soc. Am. 25: 102–104.

Kaston, B. J. 1936. The native elm bark beetle, *Hylurgopinus rufipes* Eichhoff, in Connecticut. Conn. Agric. Exp. Stn. Bull. 420, 39 pp., illus.

Kinzer, G. W., A. F. Fentiman, T. F. Page, Jr., R. L. Foltz, J. P. Vite, and G. B. Pitman. 1969. Bark beetle attractants: Identification, synthesis and field bioassay of a new compound isolated from Dendroctonus. Nature 221: 447–478.

Michael, R. R. and J. A. Rudinsky. 1972. Sound production in Scolytidae: specificity in male *Dendroctonus* beetles. J. Insect. Physiol. 18: 2189–2201.

Payne, T. L., A. A. Moeck, C. D. Willson, R. N. Coulson, and W. J. Humphreys. 1973. Bark beetle olfaction—II. Antennal morphology of sixteen species of Scolytidae. Int. J. Insect Morphol. Embryol. 2: 177–192.

Rudinsky, J. A. and R. R. Michael. 1973. Sound production in Scolytidae: stridulation by female *Dendroctonus* beetles. J. Insect Physiol. 19: 689–705.

Silverstein, R. M., R. G. Brownlee, T. E. Bellas, D. L. Wood, and L. E. Browne. 1968. Brevicomin: principal sex attractant in the frass of the female western pine beetle, *Dendroctonus brevicomis*. Science 159: 889–891.